Claims

1. The use of an assay for adenylate kinase in an *in vitro* test for the effect of external conditions on the growth characteristics of bacterial cells.

2. The use according to claim 1 wherein the test is for the sensitivity of a bacteria to an antibiotic or biostatic agent.

3. The use according to claim 1 wherein the test is to assess the growth stage of the bacteria.

4. A method for determining the susceptibility of a bacteria to a reagent, which method comprises assaying for the adenylate kinase released by lysis of bacteria from a culture containing said reagent and comparing the results with those obtained from a similar adenylate kinase assay which is either of the culture prior to addition of reagent, and/or of lysed bacteria from the same culture at a different point in time and/or of lysed bacteria from a similar culture which does not contain the reagent.

5. A method according to claim 4 wherein bacteria are lysed using a chemical lytic agent.

6. A method according to claim 4 wherein the lytic agent is specific for a particular bacteria.

7. A method according to claim 6 wherein the lytic agent is a bacteriophage which infects and lyses a specific bacterial genus, species or strain.

8. A method according to claim 4 wherein bacteria are lysed using an enzyme.

9. A method according to claim 8 wherein the enzyme is bacteriolysin.

10. A method according to claim 3 wherein the bacteria are first subjected to a separation step to substantially remove any other non-target bacteria in the culture.

11. A method according to claim 10 wherein the separation is carried out using an immunocapture method.

12. A method according to claim 11 wherein the said bacteria are concentrated at a solid surface on which antibodies or the binding fragments thereof which are specific for the target bacteria are immobilized.

13. A method according to claim 4 wherein the culture further comprises a growth medium which selectively favours the said bacteria.

14. A method according to claim 4 for determining the sensitivity of a bacteria to a lytic antibiotic, said method comprising the steps of (i) separating said bacteria from other microbial species (ii) determining the extracellular adenylate kinase content of a culture of said bacteria (iii) adding the lytic antibiotic to the culture and incubating it for a period sufficient to allow the antibiotic to exert its lytic effect, and (iv) determining the extracellular adenylate kinase content of the culture to assess whether lysis has taken place.

15. A method according to claim 14 wherein in step (i), the bacteria are separated using immunocapture techniques.

16. A method according to claim 14 ~~or claim 15~~ wherein the said culture of bacteria comprises a selective growth medium which favours said bacteria.

17. A method according to claim 4 for determining the sensitivity of a bacteria to a a non-lytic antibiotic or biostatic agent, said method comprising (i) separating said bacteria from other microbial species, (ii) incubating a culture of said bacteria in the presence of said non-lytic antibiotic or biostatic agent (iii) determining whether the total adenylate kinase content of the culture increases or decreases over the period of the incubation by removing samples at spaced time periods, lysing bacteria in these samples and assaying for adenylate kinase in said samples.

18. A method according to claim 17 wherein the bacteria are lysed using a chemical lytic agent.

19. A method according to claim 17 ~~or claim 18~~ wherein in step (i), the bacteria are separated using immunocapture techniques.

20. A method according to Claim 17 ~~any one of claims 17 to 19~~ wherein the said culture of bacteria comprises a selective growth medium which favours said bacteria.

21. A method for determining the sensisitivity of a bacteria to an antibiotic or biostatic agent, said method comprising

(a) incubating a first sample of a culture of said bacteria, a second sample in the presence of said antibiotic, a third sample in the presence of a bacteriophage which will specifically lyse said target bacteria and a fourth sample in the presence of both said bacteriophage and said antibiotic;

(b) determining the adenylate kinase content of each of the first to fourth samples after culture, and

(c) determining the sensitivity or resistivity of the bacteria on the basis of the adenylate kinase assay results and on the mode of action of the antibiotic or biostatic agent.

22. A method according to claim 21 wherein the results obtained in step (c) are compared with the results given in Figure 4 herein to determine whether the bacteria is resistant or susceptible to the antibiotic or biostatic agent.

23. A method according to claim 21 ~~or claim 22~~ wherein concentration of the said bacteria in the culture is increased prior to step (a) by an immunocapture procedure.

24. A method according to ~~any one of claims 21 to 23~~ claim 21 wherein the said samples further comprise a selective growth medium which favours growth of said bacteria in preference to other microbial species.

25. A method of determining the growth phase of a bacterial culture which method comprises

(a) subjecting a first sample of said bacterial culture to a lytic reagent so as to lyse bacterial cells therein,

(b) assaying for adenylate kinase in said first sample,

(c) assaying for adenylate kinase in a second sample of said culture which has not been exposed to the lytic agent; and

(c) comparing the results obtained from said first and second cultures and assessing the growth stage of the culture.

26. A method according to claim 25 wherein in step (c), results showing that adenylate kinase levels in the second samples which are of the order of 1% of the levels found in the first sample is indicative of a healthy, log phase culture and levels in excess of 1% are indicative of a progression into stationary phase.

27. A method according to claim 25 ~~or claim 26~~ wherein the lytic agent comprises a detergent or a bacteriophage which specifically infects and lyses particular target bacterial cells.

28. A test kit for testing the sensitivity of bacteria to antibiotics, said kit comprising one or more antibiotics, and one or more reagents necessary for assaying for adenylate kinase.

29. A test kit according to claim 28 wherein the said reagents necessary for assaying for adenylate kinase comprise ADP, a source of magnesium ions, luciferin and luciferase.

30. A test kit according to claim 28 ~~or claim 29~~ wherein the antibiotics are freeze-dried.

31. A test kit according to ~~any one of claims 28 to 30~~ Claim 28 which further comprises a lytic agent.

32. A test kit according to claim 31 wherein the lytic agent comprises a chemical agent.

33. A test kit according to claim 31 wherein the lytic agent comprises a bacteriophage which is specific for the particular target bacterial cells.

34. A test kit according to ~~any one of claims 28 to 33~~ Claim 28 which further comprises a multi-well plate.

35. The use according to claim 1 substantially as hereinbefore described with reference to the Examples.

36. A method according to claim 4 substantially as hereinbefore described with reference to the Examples.

37. A test kit substantially as hereinbefore described with reference to Example 6.